Maximizing Photosynthetic Efficiencies and Hydrogen Production in Microalgal Cultures

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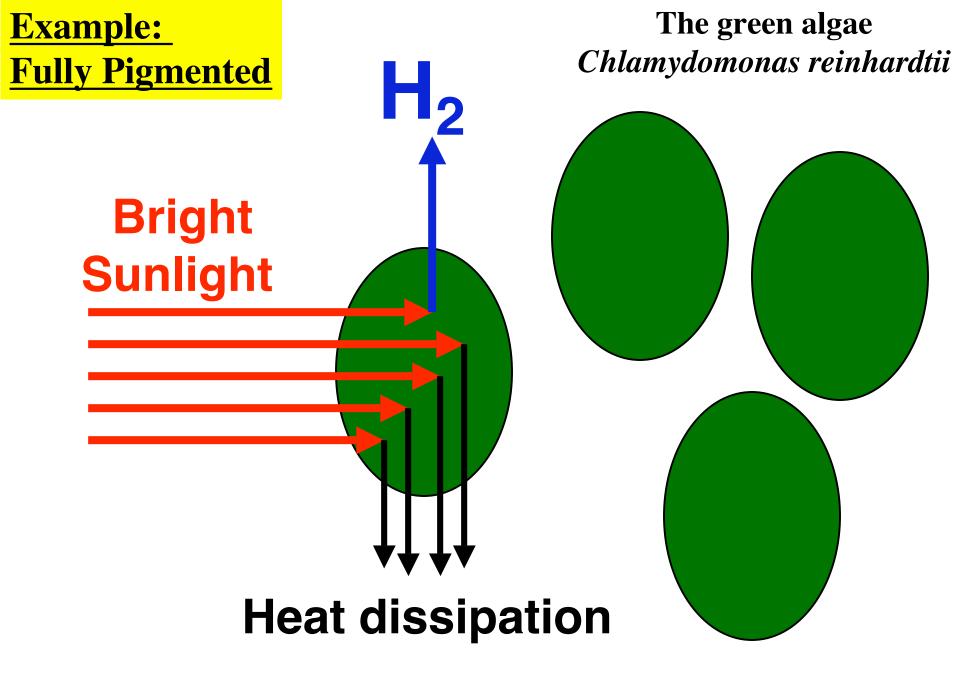
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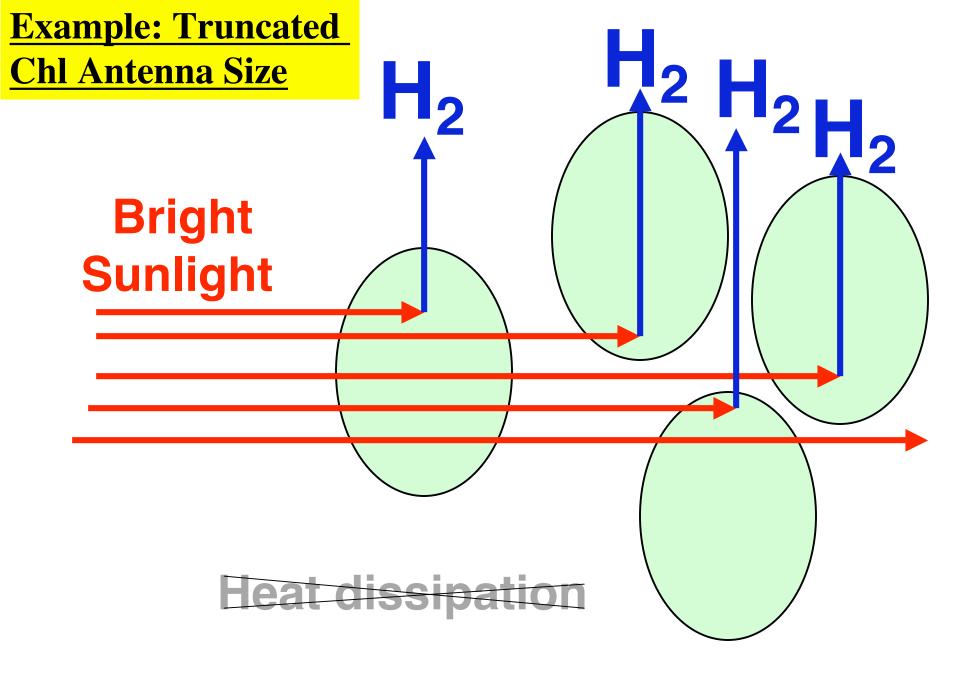
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This presentation does not contain any proprietary or confidential information

Objectives and Approach

- General Objective: Minimize the chlorophyll antenna size of photosynthesis to maximize light energy conversion efficiency in green algae.
- Approach: Employ DNA insertional mutagenesis and high-throughput screening methods to select tagged green algae with a smaller Chl antenna size.
- Ancillary Objective: Identify and characterize genes that regulate the Chl antenna size in Chlamydomonas reinhardtii.
- 2004 Target: Achieve a 7.5% Utilization Efficiency of Absorbed Light Energy in green algal photosynthesis.





Benefits from this Project

Reducing the Chl antenna size of photosynthesis is needed for any effective use of microalgae in:

- H₂ production,
- carbon sequestration,
- biomass accumulation,
- waste water treatment,
- other bio-fuels generation.

Budget - FY 2004

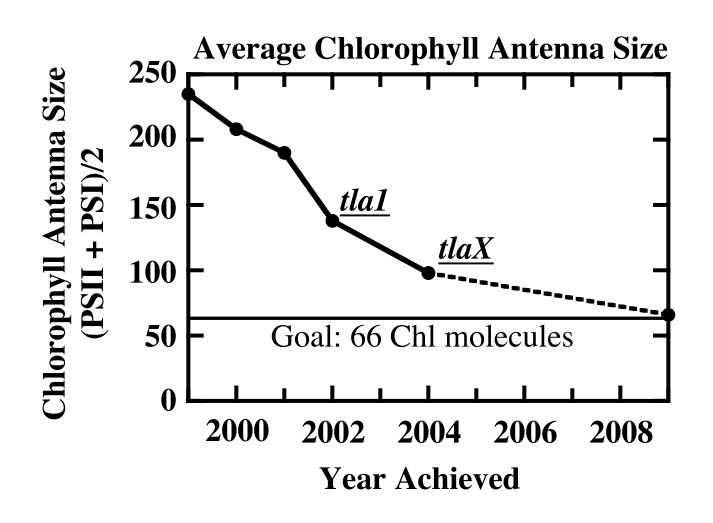
- Total DOE: \$ 200,000
- Direct: \$ 131,200
- Indirect: \$ 68,800 (Overhead)

• Cost Share (UC Berkeley): \$ 50,000

Technical Barriers and Targets

- <u>Barrier</u>: Low Light Utilization Efficiency in Photobiological Hydrogen Production due to a Large Photosystem Chlorophyll Antenna Size (Barrier I).
- <u>Topic</u>: Topic 2 (Photolytic Processes), Sub-Topic 2B (Photobiological processes), Light Utilization Efficiency problem.
- <u>Target for 2004</u>: Reach a 7.5% Utilization Efficiency of Absorbed Light Energy.

Project Timeline



Technical Accomplishments

Utilization Efficiency of Absorbed Light Energy Target for 2004: 7.5%

- Wild type antenna size = 235 Chl molecules (100%)
 (PSII=230; PSI=240)
 Photon use efficiency of WT photosynthesis = ~10%
 Utilization Efficiency of Absorbed Light Energy by WT: ~5%
- tla1 antenna size = 138 Chl molecules (59% of control)
 (PSII=115; PSI=160)
 Photon use efficiency of tla1 photosynthesis = ~20%
 Utilization Efficiency of Absorbed Light Energy by tla1: ~10%

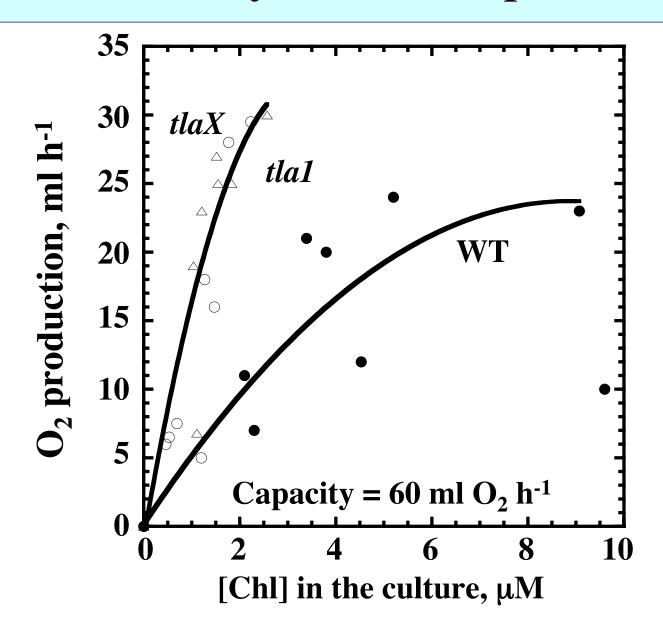
2004 Year Accomplishment

- tlaX antenna size = 98 Chl molecules (42% of control)
 (PSII=80; PSI=115)
 Photon use efficiency of tlaX photosynthesis = ~30%
 Utilization Efficiency of Absorbed Light Energy by tlaX: ~15%
- Long-term goal: 66 Chl molecules (28% of control)
 (PSII=37; PSI=95)
 Photon use efficiency of photosynthesis goal = ~60%
 Utilization Efficiency of Absorbed Light Energy goal: ~30%

Measurement in Scale-up Cultures



Productivity in Scale-up Cultures



Current State of the Art

- Significant progress and ahead-of-schedule timeline in terms of acquiring "truncated Chl antenna size" mutants. *This demonstrates feasibility and suitability of the approach.*
- Have completed characterization of the role of *Lhcb* and *CAO* gene expression in the regulation of the Chl antenna size.
- Have not yet completed characterization of the *Tla1* and *TlaX* genes, neither do we know the mode by which these novel genes function in the regulation of the Chl antenna size in photosynthetic organisms.

Interactions and Collaborations

- Collaboration with NREL and ORNL (Made available to NREL and ORNL the *tlaX* truncated Chl antenna mutant for use in their Photobiological Hydrogen Production project.)
- Interactions with the Chrysler Corporation (Recipient of a Chrysler "University Research Opportunity Award". Advising the Technical Affairs division of DaimlerChrysler on matters of Hydrogen Biotechnology.)

Responses to Previous Year Reviewers Comments

• Is automated lab equipment available that would be of significant help in moving project forward?

My lab is well provided with automated equipment for the **Chl antenna size** analyses (sole source), as well as for the conduct of biochemistry, biophysics and molecular genetics RD&D. Moreover, UC Berkeley operates specialized facilities (automated **DNA sequencing, polyclonal antibody generation, microscopic imaging, greenhouses** etc.). These subsidized facilities serve to support research efforts on campus. In addition, this project in my lab further benefits from the recent sequencing of the *Chlamydomonas reinhardtii* genome by the **DOE's Joint Genome Institute** in nearby Walnut Creek, CA.

• Is cost and effectiveness easily justified?

In addition to the direct cost sharing, this project is further supported by the University in the form of **relatively low overhead and subsidized facilities**. This is possible because of the **instructional and training mission** of this public institution. As a result, progress is achieved at a fraction of the cost that would be required by government laboratories or industry.

Future Work

Remainder of FY 2004

1. Advance the biochemical and molecular characterization of the *tlaX* strain. Publish *tla1*- and *tlaX*-related analyses.

FY 2005 and Beyond

- 1. Functionally characterize the corresponding *tla1* and *tlaX* genes (how do they work?)
- 2. Establish transformation (sense and antisense) protocols with *Tla*-type genes to further down-regulate the Chl antenna size in *Chlamydomonas reinhardtii*.
- 3. Perform comparative green-alga light utilization efficiency and photosynthetic (H_2) productivity measurements under mass culture conditions in wild type and Tla-type mutants.
- 4. Perform genetic crosses to combine different *tla*-type properties.

Safety Aspects

- Identification and discussion of safety vulnerability techniques used in the analysis of the design and operation of equipment for this project: Pressurized cylinders with hydrogen, helium and argon that are employed in the conduct of this work are safely anchored in appropriately designed berth spaces.
- Identification of management of change process used for the project: Training in general, and specific aspects of safety for this project, is mandatory for all employees in this department. The small amounts of H₂ involved in this work do not entail any special precautions.
- Other safety-related insights benefiting the project and/or of potential application to other projects, e.g. experiences with management of change (MOC) procedures: None